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Patent Pending

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Auro•Dex® Helicobacter pylori MultiTest

Test for the Rapid Detection of Helicobacter pylori IgG, IgA, & IgM Antibodies in Human Serum.

For In Vitro Use Only

INTENDED USE

The intended use of the Auro•Dex® *H.pylori* MultiTest is for the rapid detection of *Helicobacter pylori*-specific antibodies in human serum.

BACKGROUND

Helicobacter pylori formerly known as *Campylobacter pyloridis* or *C.pylori,* has been more recently implicated in the pathogenesis of gastric and duodenal ulcers and a variety of other non-ulcerative gastric diseases (3, 4). *H.pylori* first cultured from gastric biopsies of ulcer sufferers, by Warren and Marshall, was later characterized as a gram negative, micro aerobic multi-flagellate, urease producing microorganism, often associated with gastric and duodenal ulcerations (5).

It has been reported that *H.pylori* is present in over 80% of symptomatic patients diagnosed with duodenal and gastric ulcers (6). It has also been reported that successful therapy is associated with marked clinical improvement and eradication of *H.pylori,* further strengthening the evidence linking *H.pylori* to gastrointestinal ulcerative diseases (7).

The immune response to *Helicobacter pylori* infection is the development of *H.pylori* specific antibodies (8). Because of the high prevalence of *H.pylori* in the general population, low antibody titers can be also found in asymptomatic individuals (9). However, chronic and active infections are likely associated with the development of high titers of *H.pylori* specific antibodies (10). Therefore, as an aid in the diagnosis of gastrointestinal ulcerative diseases, it is helpful to test for the presence of antibodies to *H.pylori*.

To date, several strategies using antimicrobial agents have proved effective in eradicating H.pylori and preventing strain resistance. Quadruple therapy is currently the regimen of choice for treating most patients (17, 18).

PRINCIPLE OF THE TEST

The Auro•Dex® *H.pylori* MultiTest is a lateral flow, immunochromato-graphic screening test. In the assay, the test serum is allowed to react with *H.pylori* antigen conjugated to gold particles. The mixture then flows laterally on the membrane to a test region (G, A, M) where immobilized antibodies (anti-IgG, anti-IgA, anti-IgM) capture the serum antibody-gold conjugated *H.pylori* complex. A red colored line formed in the test region indicates the presence and class of *H.pylori* antibodies in the test specimen. Absence of the colored line in the test region indicates a negative result. A red colored line in the control region (C) will always appear regardless of the presence or absence of *H.pylori* specific antibodies in tested serum.

PACKAGING

The packing consists of: - individually sealed test devices
- disposable plastic pipettes
- 1 dropper tip vial containing buffer

STORAGE AND STABILITY

1. Store test device in its sealed pouch refrigerated (2ºC to 8ºC) or at room temperature. **DO NOT FREEZE**.

2. Do not use beyond expiration date.

SPECIMEN COLLECTION AND HANDLING

No special patient preparation is necessary. Blood should be collected by accepted medical practice. Serum should be separated by appropriate technique. No additives or preservatives need to be added. Store serum samples refrigerated at 2ºC to 8ºC. For storage longer than 10 days, freeze samples at -20ºC.

PRECAUTIONS AND WARNINGS

1. HANDLE ALL ASSAY SPECIMENS AS CAPABLE OF TRANSMITTING INFECTIOUS AGENTS.

2. FOR IN VITRO USE ONLY.

3. As with any possible infectious materials, proper laboratory procedures should be followed and precautions must be taken.

4. DISPOSAL. All test material should be disposed of by appropriate means used for medical/biological waste.

ASSAY PROCEDURE

1. Bring all components to room temperature. Remove the device and the plastic pipette by tearing open the pouch. Label the device with patient's name and place device horizontally on a working bench.

2. a. If testing one sample at a time, no pre-dilution is required.

   - With the plastic pipette supplied, add 1 drop of serum/plasma into the sample well(s).

   - **IMMEDIATELY** (within 5 seconds) after sample addition, add 2 drops of buffer into sample well(s).

   OR

   b. If multiple tests are run simultaneously, pre-dilution of sample is required because it is otherwise difficult to add buffer immediately.

   - Holding the buffer dropper tip bottle vertically, carefully add 2 drops of buffer into an empty sample tube.

   - With the plastic pipette supplied, add 1 drop of serum/plasma into the sample tube. Mix by swirling and add all contents to the device sample well(s).

3. Within 2 - 5 minutes observe the development of a red indicator red line beside the C (control) and potential red lines beside the letters G, A, M (tests). Read and interpret the results within 8 - 10 minutes, but not longer than 15 minutes (see diagrams).

INTERPRETATION OF THE RESULTS

Positive: Presence of the control (C) line, plus either: G only, A only, G, A and M, G and M, G and A or A and M indicates class specific antibodies to H.pylori.

Negative: Presence of only a single red line in the control region (C) indicates absence of antibodies to *H.pylori* in the test specimen.

NOTE: If after 15 minutes no red control line is visible within the test window, the test should be repeated with a new device. Do not interpret results after 15 minutes.

The presence of IgM only, should be considered an indeterminate result, and should be followed by repeat testing at two to four weeks. (see below)

CLINICAL SIGNIFICANCE of IgG, IgA, and IgM ANTIBODIES to H. PYLORI

Several studies have shown the importance of determining the immunoglobulin class reactivity to *H.pylori*. (11,12,13). Elevated levels of serum immunoglobulin IgA anti-*H.pylori* have been found to be associated with an increased risk of gastric cancer, especially when combined with low serum pepsinogen levels (14). Decreasing levels of *H.pylori* IgG, IgA and IgM antibodies are a reliable indicator of bacterial eradication after antimicrobial treatment (15). While 97% of successfully treated patients exhibited a significant decrease in IgG, IgA and IgM titres, patients who remained infected with *H.pylori* showed unchanged or rising titres of IgG, IgA and IgM. Increased IgM anti-*H.pylori* combined with weak or elevated IgG anti-*H.pylori* may be indicative of a chronic *H.pylori* infection (16).

LIMITATION OF THE ASSAY

1. Result of the test should be used as an aid in diagnosis and should be interpreted in light of all clinical findings and diagnostic procedures.

2. The test procedure and interpretation of the results must be followed closely to obtain reliable results.

3. The test device is for one patient, single use only. DO NOT REUSE.

PERFORMANCE CHARACTERISTICS

The performance of the Auro•Dex® *H.pylori* lateral flow test was compared to two commercially available EIA kits. The sensitivity and specificity of the Auro•Dex® test in detecting antibodies to *H.pylori* were: sensitivity 95.9%, specificity 97.4% and an overall correlation of 97.0%.